MagnaBioSciences, LLC
a Quantum Design International company

MICT® FSH

Quantitative FSH Test in Human Serum

CE IVD REF **850002**

INTENDED USE

MICT FSH is an *in vitro* diagnostic test for the quantitative measurement of follicle stimulating hormone (FSH) levels in human serum using the MICT System.

INTRODUCTION

Follicle stimulating hormone (FSH) is a glycoprotein with a molecular weight of approximately 30KD. It is produced by the pituitary gland in response to gonadotropin-releasing hormone (GnRH), secreted by the hypothalamus [1]. FSH is a heterodimer composed of two different sub-units, alpha (α) and beta (β).The α sub-unit contains 89 amino acids, is common to other gonadotropins, such as LH, hCG and TSH. The β sub-unit, contains 118 amino acids, is specific for FSH and confers its biochemical specificity [1].

FSH stimulates the growth and maturation of the ovarian follicles in women, spermatogenesis and the maturation of germ cells in men [2]. Immunoassays for serum levels of FSH, and for luteinizing hormone (LH), are useful in the evaluation of disorders of reproduction and puberty, such as hypogonadism, ovulation irregularity, and infertility. In addition, serum levels of FSH and LH are monitored in ovulation induction and in the clinical administration of gonadotropins [3, 4].

PRINCIPLE OF THE TEST

MICT FSH is a magnetic immuno-chromatographic test (MICT) using Super Para-magnetic Particles (SPMPs) as the labeling agent. The SPMP magnetic field is quantified by a MICT® Instrument.

MICT FSH is based on the principle of capillary flow and separation of immune complexes bound to SPMPs within a porous matrix. In the test device, FSH antibody (Ab) - antigen (Ag) immune reactions occur on a narrow strip of capillary membrane. The narrow strip is housed in a disposable plastic cassette to form a test device.

The specimen containing the FSH antigens is mixed with a lyophilized conjugate pellet and added into the sample well of the MICT cassette. As the FSH Ab-Ag-SPMP immune complexes migrate through the test strip, they are captured by the second antibodies at the test line. A Control line is formed by the control SPMP immune complexes. The MICT instrument is used to analyze the quantity of magnetic particles captured on both control and test lines. A visible line is not necessary for interpreting the MICT test result. The FSH concentration is directly proportional to the magnetic signal and is reported based on the predetermined test algorithms that are embedded in the cassette barcode labels.

REAGENTS AND EQUIPMENT

Reagents and materials supplied:

- MICT FSH Device packs (20)
  Each pack contains (1) MICT FSH cassette, which contains murine monoclonal anti-FSH antibodies (test line) and control goat-anti-rabbit IgG polyclonal antibodies (control line); and (1) lyophilized FSH antibody conjugate tube, which contains anti-FSH monoclonal antibodies covalently coupled to the SPMPs.

Materials required but not provided

MICT Instrument
100µl mini-pipette
Serum collection tube
Centrifuge
Timer

STORAGE
Store at 1 – 30 °C

PRECAUTIONS
- The test is designed for *in vitro* diagnostics and is for professional use only.
- Wear protective gloves and masks throughout the test procedure. Wash hands thoroughly after handling specimens.
- Do not pipette by mouth. Do not eat, drink or smoke in designated work areas.
- Follow standard guidelines for proper handling and disposal of patient specimens, chemical reagents, used cassettes and all other materials used during the test.
- MICT® tests are stable within the expiration date when stored and handled as directed. Do not use MICT® tests beyond the expiration date.
- Do not use a cassette from a compromised pouch.
- Specimens with repeated freeze-thaw cycles can cause abnormal results.

SPECIMEN COLLECTION AND STORAGE
For handling, processing and storing blood samples, read the following notes, as well as the manufacturer's procedures on sample collection tubes.
- Follow routine procedures and precautions for venipuncture.
- Draw blood into serum collection tubes. Keep the tubes sealed at all times.
- For serum samples, allow blood to clot completely before centrifugation.
- Samples should be centrifuged and refrigerated within 2 hours of blood draw.
- Serum should be physically separated from cells as soon as possible, with a maximum time limit of 2 hours from centrifugation. Remove any residual fibrin or cellular matter.
- Turbid serum samples containing particular matter should be transferred from the original tube and re-centrifuged prior to assay.
- Samples should be tested within 8 hours of blood draw.

ASSAY PROCEDURE
- Open the foil pouch; take out the conjugate tube and the test cassette. Place the cassette on a dry, flat surface. The cassette should be used immediately after opening the pouch. Mark patient ID on the cassette as necessary.
- Hold the conjugate tube or place it in a rack. Make sure that the small conjugate pellet is at the bottom of the tube by gently tapping the wall of the tube.
- Use a standard 100 µL pipette and add 100 µL of patient serum sample into the conjugate tube. Mix the serum with the lyophilized conjugate pellet by pipetting up and down 3 - 4 times.
- Use the same pipette tip from the previous step, transfer the entire contents of the mixed solution into the sample well on the test cassette. Set a timer and wait for 30 minutes.
- After the 30 minutes, insert the test cassette into the MICT instrument and close the door. The instrument will immediately initiate the barcode reading and perform test verification. In approximately 10 - 15 seconds, the verification will be complete as indicated by the illuminated green "READY" light.
- Verify test name and sample source displayed on the instrument screen, input patient ID and/or patient number, and then press the "START" button. This initiates the MICT analysis of the test and control lines. The FSH serum concentration in mIU/mL is displayed and printed.
- For detailed instructions on how to use the MICT instrument, refer to the MICT Instrument User Manual.

RESULTS
The MICT instrument automatically determines FSH concentration in the sample using the information on the cassette barcode label.

CALIBRATION
A calibration master curve is generated for each lot of MICT FSH, using master calibrators referenced to the WHO 2nd international standard for human FSH (IS 94/632). The master curve information is transferred to the MICT instrument through the barcode label on the test cassette. The MICT TSH assay requires no calibration at the customer site.

QUALITY CONTROL

A. MICT Instrument Verification

The performance of the MICT instrument can be tested on a regular basis using the verification cassette (supplied with the MICT instrument). The verification cassette contains three test regions on the strip that are pre-measured and are stable magnetic signals. These signals are used to verify the instrument internal calibration and system performance. After inserting the verification cassette, the MICT instrument will perform a series of measurements and compare the results to the calibrated values stored in the test device barcode. Verification results should pass on all test items including the Overall Test, Accuracy and Noise.

B. Built-in Procedural Control

A built-in control line is present to assure that 1) reagents and test devices are functional; 2) test procedures are carried out correctly; and 3) test results are valid. If the control line value does not meet the specific QC standard, an error message will appear, indicating that the test is invalid. If this occurs, the test must be repeated.

C. FSH Controls

Good laboratory practice including the use of control materials should be followed to ensure the test performance. Follow the federal, state, and local requirements for each individual laboratory. If FSH controls are out of range, re-run the test. If the problem continues, contact MagnaBioSciences or your local distributors for technical support.

LIMITATION OF PROCEDURE

- The performance of the MICT FSH has been established using serum samples only.
- Specimens can be measured within the reportable range. For accurate results, serum FSH levels greater than 100 mIU/mL should be tested with another method.
- Specimens from patients who have been exposed to mouse monoclonal antibodies may interfere with the test results, and show falsely elevated or depressed FSH values.
- For diagnostic purposes, the MICT FSH results should always be assessed in conjunction with other diagnostic findings and information obtained by the physician. If the FSH level is inconsistent with clinical evidence, additional FSH testing is suggested to confirm the result.
- MICT FSH immunoassay does not show a high-dose hook effect up to 5,000 mIU/mL

EXPECTED RANGE

Expected values in serum for FSH are summarized in the table below[5]:

	FSH mIU/mL
Males 23 -70 yrs	1.4 – 15.4
Females	
Follicular Phase	1.4 – 9.9
Midcycle Peak	0.2 – 17.2
Luteal Phase	1.1 – 9.2
Postmenopausal	19.3 – 100.6

PERFORMANCE CHARACTERISTICS

Precision

The reproducibility of MICT FSH was measured by duplicate testing of pooled serum samples (n = 60) over a ten-day period using two MICT Systems and two lots of MICT FSH tests. The test precision data are summarized below.

MICT FSH Precision (CV%)

Sample	Mean [FSH] (mIU/mL)	Between Run CV%	Between Analyzer CV%	Between Reagent Lot CV%	TOTAL CV%
1	13.5	10.0	1.0	4.0	10.8
2	45.6	9.0	2.0	5.0	10.5
3	84.6	7.0	3.0	4.0	8.6

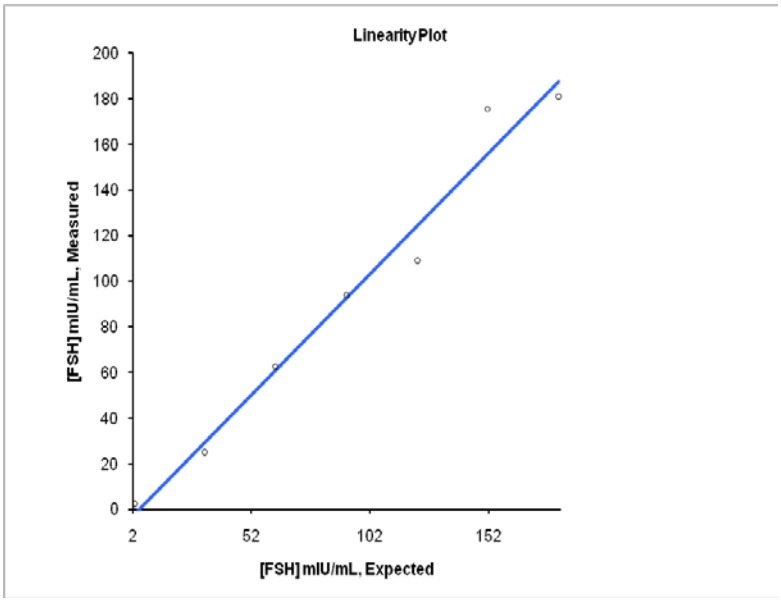
Method Comparison

One hundred and twenty-three (123) clinical samples were used to compare the serum values measured by the MICT FSH and the Siemens ADVIA Centaur FSH immunoassay. The agreement of the two methods was evaluated using Demings regression analysis. Pearson correlation r-value was used to test association between the values.

n	Range of Observations (mIU/mL)	Intercept (mIU/mL)	Slope	r
123	0.3 - 128	-3.19	1.09	0.91

Linearity

Linearity was determined in accordance with CLSI protocol EP6-A. High and low value patient serum samples were used to prepare a set of linearity samples. The two samples were mixed in different ratios and the FSH values determined using the MICT system and the results compared them with expected values. The graph below summarizes the data.



Interfering Substances

Interfering substances were added to serum samples containing known amounts of FSH. The value obtained for the serum with each interfering substance was compared to the value obtained for the serum without the interfering substance. These additives did not show interference at the levels indicated.

Additives	Concentration
Bilirubin	20 mg/dL
Hemoglobin	300 mg/dL
Triglycerides	1,000 mg/dL

Sensitivity and Assay Range

Two lots of the MICT FSH test were evaluated for sensitivity using the CLSI EP17-A protocol. The LOB, LOD, and LOQ are summarized as the average obtained from the two lots.

Sensitivity Parameter	mIU/mL
Limit of Blank (LOB)	0.01
Limit of Detection (LOD)	0.12
Limit of Quantitation (LOQ)	0.22

The assay range is 0.22 mIU/mL to 100 mIU/mL

Specificity

The antibodies used in the MICT FSH were tested for potential cross-reactivity against the closely related hormones: TSH, hCG and LH. The results are summarized below.

Hormone	Concentration	Cross-Reactivity (%)
TSH	2 mIU/mL	nd
hCG	200K mIU/mL	<0.2
LH	3000 mIU/mL	nd

REFERENCES
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powered by MAR.

The MICT® measurement technology and assays are covered by US Patents 6,046,585; 6,275,031; 6,437,563; 6,483,303; 6,518,747; 6,597,176; 6,607,922; 6,927,570 and foreign equivalents. Other patents are pending.